

WE CLAIM:

1. A composition for stimulating an immune system, said composition comprising a plurality of fused cells, each of which fused cells is generated by fusion between at least one mammalian dendritic cell and at least one mammalian non-dendritic cell that expresses a cell-surface antigen, wherein at least half of the fused cells express, in an amount effective to stimulate an immune system, (a) a MHC class II molecule, (b) B7, and (C) the cell-surface antigen.

5 2. The composition of claim 1, wherein the mammalian non-dendritic cell is a cancer cell.

10 3. The composition of claim 1, wherein the mammalian dendritic cell and the mammalian non-dendritic cell are obtained from the same individual.

4. The composition of claim 3, wherein the individual is a human.

5. The composition of claim 4, wherein the cell-surface antigen is a cancer antigen.

20 6. The composition of claim 1, wherein the mammalian dendritic cell and the mammalian non-dendritic cell are obtained from different individuals of the same species.

7. The composition of claim 6, wherein the species is *Homo sapiens*.

25 8. The composition of claim 7, wherein the cell-surface antigen is a cancer cell antigen.

9. A method of producing a fused cell useful for stimulating an immune system, comprising:

30 providing a first fused cell formed by fusion between at least one mammalian dendritic cell and at least one mammalian non-dendritic cell that expresses a cell-surface antigen; and fusing the first fused cell with at least one mammalian dendritic cell to produce a second fused cell that is useful for stimulating an immune system.

10. The method of claim 9, wherein the second fused cell expresses (i) a MHC class II molecule, (ii) B7, and (iii) the cell-surface antigen.
- 5 11. The method of claim 9 wherein all of the mammalian dendritic cells and the mammalian non-dendritic cells are human cells.
- 10 12. The method of claim 11, wherein the cell surface antigen is a cancer antigen.
- 15 13. A method of producing a fused cell, comprising:
providing a cell sample comprising (i) a first plurality of mammalian dendritic cells, and
(ii) a plurality of mammalian non-dendritic cells expressing a cell-surface antigen; and
contacting the cell sample with a fusion agent to produce a post-fusion population of cells comprising a fused cell that is the fusion product of at least one of the dendritic cells and at least one of the non-dendritic cells.
- 20 14. The method of claim 13, wherein the fused cell is useful for stimulating an immune system.
- 25 15. The method of claim 14, wherein the fused cell expresses (a) a MHC class II molecule, (b) B7, and (C) the cell-surface antigen.
- 30 16. The method of claim 13, wherein the mammalian dendritic cells are cultured from (i) bone marrow cells, or (ii) peripheral blood cells.
17. The method of claim 16, wherein the time between the contacting step and the separating step is less than 10 days.
18. The method of claim 13, further comprising fusing the isolated fused cell with at least one mammalian dendritic cell to produce a secondary fused cell.
19. The method of claim 18, wherein the secondary fused cell expresses (i) a MHC class II molecule, (ii) B7, and (iii) the cell-surface antigen.

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20. The method of claim 19, wherein all of the mammalian dendritic cells and the mammalian non-dendritic cells are human cells.

21. The method of claim 20, wherein the cell surface antigen is a cancer antigen.

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22. A method of stimulating the immune system in an individual, said method comprising administering the composition of claim 1 to the individual.

23. The method of claim 22, wherein the individual has a condition selected from the group consisting of:

susceptibility to infection with an intracellular pathogen;
infection with an intracellular pathogen;
cancer; and
predisposition to develop cancer.

24. A method of stimulating the immune system in a human, said method comprising administering the composition of claim 1 to the human.

25. The method of claim 24, wherein the mammalian dendritic cells are obtained from the human or an identical twin of the human.

26. The method of claim 25, wherein the non-dendritic cells are cancer cells obtained from the human.

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27. The method of claim 25, wherein the cell-surface antigen is a cancer antigen.

28. The method of claim 25, wherein the cell-surface antigen is an antigen derived from a pathogen.

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29. The method of claim 28, wherein the pathogen is a virus.

30. The method of claim 27, wherein the cancer antigen is MUC1.

31. The method of claim 30, wherein the individual has one of the following conditions or predisposition to develop one of the following conditions: breast cancer, ovarian cancer, pancreatic cancer, prostate gland cancer, lung cancer and myeloma.

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J6a/1 >* 32. A substantially pure population of educated, antigen-specific immune effector cells expanded in culture at the expense of hybrid cells, wherein the hybrid cells comprise antigen presenting cells fused to cells that express one or more antigens.

10 33. The population according to claim 32, wherein the antigen presenting cells are dendritic cells.

34. The population according to claim 32, wherein the cells expressing the antigen(s) are tumor-specific.

15 J6c/2 > 35. The population according to claim 32, wherein the antigen-specific immune effector cells are cytotoxic T lymphocytes.

36. The population according to claim 32, wherein the antigen-specific immune effector cells are genetically modified cells.

20 37. The population according to claim 32, wherein the hybrid cells are genetically modified cells.

25 38. The population according to claim 36, wherein the genetic modification comprises introduction of a polynucleotide.

39. The population according to claim 38, wherein the polynucleotide encodes a peptide, a ribozyme or an antisense sequence.

30 40. The population according to claim 32, wherein the antigen presenting cells and the cells that express one or more antigens are autologous.

41. The population according to claim 32, wherein the antigen presenting cells and the cells that express one or more antigens are allogeneic.

Sule C3 42. A substantially pure population of educated, antigen-specific immune effector cells produced by culturing immune effector cells with hybrid cells, wherein the hybrid cells are antigen presenting cells fused to cells that express one or more antigens and wherein the educated, antigen-specific immune effector cells are expanded at the expense of the hybrid cells.

5 43. The population according to claim 42, wherein the antigen presenting cells are dendritic cells.

10 44. The population according to claim 42, wherein the cells recognizing the antigen(s) are tumor-specific.

Sule C4 45. The population according to claim 42, wherein the antigen-specific immune effector cells are cytotoxic T lymphocytes.

t5 46. The population according to claim 42, wherein the antigen-specific immune effector cells are genetically modified cells.

20 47. The population according to claim 42, wherein the hybrid cells are genetically modified cells.

48. The population according to claim 46, wherein the genetic modification comprises introduction of a polynucleotide.

25 49. The population according to claim 48, wherein the polynucleotide encodes a peptide, a ribozyme or an antisense sequence.

50. The population according to claim 42, wherein the antigen presenting cells and the cells that express one or more antigens are autologous.

30 51. The population according to claim 42, wherein the antigen presenting cells and the cells that express one or more antigens are allogeneic.

Sule B5 52. The population according to claim 42, wherein the immune effector cells are

naïve.

53. The population according to claim 42, wherein the immune effector cells are educated.

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54. The population according to claim 42, wherein the immune effector cells are produced by culturing immune effector cells with hybrid cells in the presence of a cytokine.

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55. The population of claim 54, wherein the cytokine is IL-2.

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56. A method for producing antigen-specific immune effector cells comprising culturing immune effector cells in an effective amount of hybrid cells, wherein the hybrid cells comprise antigen presenting cells fused to cells expressing one or more antigens and wherein the antigen-specific immune effector cells are produced at the expense of the hybrid cells.

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57. The method according to claim 56, wherein the antigen presenting cells are dendritic cells.

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58. The method according to claim 56, wherein the dendritic cells are derived from blood, bone marrow or skin and the immune effector cells are derived from tumor tissue.

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59. The method according to claim 56, wherein the cells expressing the antigen(s) and the immune effector cells have been enriched from a tumor.

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60. The method according to claim 56, wherein the antigen presenting cells and the cells that express one or more antigens are autologous.

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61. The method according to claim 56, wherein the antigen presenting cells and the cells that express one or more antigens are allogeneic.

62. The method according to claim 56, wherein the immune effector cells are naïve.

63. The method according to claim 56, wherein the immune effector cells are

educated.

64. The method according to claim 56, further comprising culturing the immune effector cells in the presence of an effective amount of cytokine.

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65. The method according to claim 64, wherein the cytokine is IL2.

66. A method of adoptive immunotherapy, comprising administering to a subject a population of educated, antigen-specific immune effector cells expanded in culture at the expense of hybrid cells, wherein the hybrid cells comprise antigen presenting cells fused to cells that express one or more antigens.

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67. A method of adoptive immunotherapy comprising administering to a subject a population of educated, antigen-specific immune effector cells made by culturing naïve immune effete cells with hybrid cells, wherein the hybrid cells are antigen presenting cells fused to cells that express one or more antigens and wherein the educated, antigen-specific immune effector cells are expanded at the expense of the hybrid cells.

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68. The method according to claim 66 or 67, wherein the antigen presenting cells are dendritic cells.

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69. The method according to claim 66 or 67, wherein the dendritic cells are derived from blood, bone marrow or skin and the immune effector cells are derived from a tumor.

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70. The method according to claim 66 or 67, wherein the cells that express one or more antigens and the immune effector cells have been enriched from a tumor.

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71. The method according to claim 66 or 67, wherein the immune effector cells are cytotoxic T cells.

72. The method according to claim 66 or 67, wherein the antigen specific immune effector cells administered to the subject are allogeneic.

73. The method according to claim 66 or 67, wherein the antigen specific immune

effector cells administered to the subject are autologous.

74. The method according to claim 66 or 67, further comprising culturing the immune effector cells in the presence of an effective amount of a cytokine.

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75. The method according to claim 74, wherein the cytokine is IL2.

76. A method of identifying a fragment of a gene encoding an antigen recognized by the population of antigen-specific immune effector cells according to claim 32 or 42, the method comprising:

- (a) providing a population of first cells of claim 32 or 42, wherein the cells have an identified major histocompatibility complex (MHC) restriction and one or more second cells having a compatible major histocompatibility complex (MHC) to the first cell but which does not express antigen;
- (b) identifying polynucleotides encoding a peptide sequence motif in the antigen displayed by the population of first cells of claim 32 or 42;
- (c) identifying polynucleotides which are aberrantly expressed by the first cells as compared to one or more second cells; and
- (d) comparing the polynucleotides identified in step (c) with the polynucleotides motifs identified in step (b) to identify the fragment of the gene encoding the antigen recognized by the immune effector cell.

77. The method of claim 76, wherein step (c) is performed prior to step (b).

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78. A vaccine comprising an antigen identified according to the method of claim 76.

79. A method of identifying a polypeptide encoding a sequence motif present in an antigen recognized by the population of antigen-specific immune effector cells according to claim 32 or 42, comprising:

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- (a) providing a cell population of antigen-specific immune effector cells of claim 32 or 42 and having an identified major histocompatibility complex (MHC) restriction; and
- (b) identifying a polypeptide encoding a sequence motif in the antigen recognized by the immune effector cells.

80. A vaccine comprising an antigen identified according to the method of claim 79.
81. A vaccine comprising a composition for stimulating an immune system according
5 to claim 1 and a pharmaceutically acceptable carrier.
82. The vaccine of claim 81 further comprising an immunoregulatory cytokine.
83. A vaccine comprising a composition for stimulating an immune system produced
10 according to the method of claim 9.
- Step 84*
84. A vaccine comprising the population of antigen-specific immune effector cells
of claim 32 or 42 and a pharmaceutically acceptable carrier.
85. A vaccine comprising antigen-specific immune effector cells produced according
to the method of claim 56.
86. A method of identifying a fragment of a gene encoding an antigen recognized by
the population of antigen-specific immune effector cells according to claim 32 or 42, the method
comprising:
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(a) providing a population of first cells of claim 32 or 42, wherein the cells
have an identified major histocompatibility complex (MHC) restriction and one or more second
cells having a compatible major histocompatibility complex (MHC) to the first cell but which
does not express antigen;
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(b) identifying polynucleotides encoding a peptide sequence motif in the
antigen displayed by the population of first cells of claim 32 or 42;
(c) identifying polynucleotides which are aberrantly expressed by the first
cells as compared to one or more second cells; and
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(d) comparing the polynucleotides identified in step (c) with the
polynucleotides motifs identified in step (b) to identify the fragment of the gene encoding the
antigen recognized by the immune effector cell
wherein the polynucleotides are identified using the SAGE method.
87. The method of claim 86, wherein step (c) is performed prior to step (b).

88. A method of identifying a polypeptide encoding a sequence motif present in an antigen recognized by the population of antigen-specific immune effector cells according to claim 32 or 42, comprising:
- (a) providing a cell population of antigen-specific immune effector cells of claim 32 or 42 and having an identified major histocompatibility complex (MHC) restriction; and
 - (b) identifying a polypeptide encoding a sequence motif in the antigen recognized by the immune effector cells wherein the polypeptide is identified using the SPHERE method.

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